Precision Sample Control and Extraction Component

Klint A. Rose
(925) 423-1926
rose38@llnl.gov

Incorporating microfluidic-based technologies into bio-analytical instruments has many benefits, including reduced sample consumption, faster response times, and improved sensitivity. To achieve these benefits, microfluidic analysis often requires sample concentration and separation techniques to isolate and detect an analyte of interest. Complex or scarce samples may also require an orthogonal separation and detection method or off-chip analysis for confirmation of results. To perform these additional steps, the concentrated sample plug must be extracted from the primary microfluidic channel with minimal loss of sample and with minimal dilution. Extracting these samples requires precise metering and control of nanoliter volumes of fluid.

Extraction of concentrated samples has been demonstrated, requiring constant control of complex electrode structures. This project reduces to practice sample extraction using droplet and bubble generation techniques. These methods are integrated with the equipment and software necessary to improve sample exchange among LLNL microfluidic technologies and improve their functionality. This technology will enable LLNL to supply the next generation biothreat detection instrumentation by improving microfluidic device performance.

Project Goals

The overall goal of this project is to deliver a universal ability to manipulate and extract small volumes of fluid (pico- to microliters) from micron-scale channels. The desired system functions include precise and reproducible control of small volumes of fluid within a microchannel, and the extraction of these sample volumes with minimal effect on the main microfluidic module's performance.

An illustration of this concept is shown in Fig. 1. These functions should be achieved in platforms with fabrication protocols that are standard in the LLNL cleanroom.

Relevance to LLNL Mission

This project addresses needs identified in LLNL's Engineering roadmaps for the detection of biomolecules (DNA, RNA and proteins) and viruses at low concentrations (10 to 1000 copies/ml). Specifically, this project addresses the necessary (and often neglected) front-end sample preparation through precise control and high-yield sample extraction of targeted analytes from upstream concentration, purification and/or separation-based microfluidic devices. This new functionality will enable additional off-chip postprocessing procedures such as DNA/RNA microarray analysis, RT-PCR, and culture growth to validate chip performance. Many LLNL programs will benefit from the improved front-end preparation offered by the precision fluidic control and sample extraction capabilities created by this project.

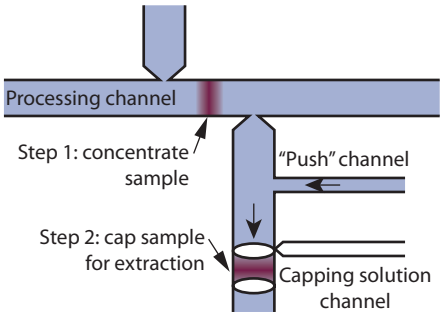

Figure 1. Flow-rate stability comparison of commercially available pump versus flow-control system created for precision sample extraction.

FY2006 Accomplishments and Results

We have accomplished our overall goal by delivering the protocols and microfluidic architecture necessary for manipulating and extracting small volumes of fluid from micron-scale channels. Specific results and accomplishments include the following:

1. Used a system for precise sample control, which includes a flow-rate sensor as feedback for a pressure unit. This system reduced the standard deviation in flow rate to 5% of that for a commercially available precision pump.
2. Created protocols for fabricating microfluidic chips for extraction. This protocol includes the hydrophobic coating necessary for capping aqueous samples. An example is shown in Fig. 2.
3. Demonstrated the capping of samples in a microfluidic channel using air bubbles and fluorinert droplets, as shown in Fig. 3. Capping with incompressible fluorinert droplets provided the most reproducible results compared to air.
4. Verified effectiveness of capping samples to eliminate loss of solute. Fluorinert-capped fluorescent samples were analyzed over thirty minutes and demonstrated no diffusion or detectable loss in concentration.
5. Demonstrated through modeling and experimental flow measurements that proposed injection of samples into fluorinert or air (inverse of capping technique) results in sample loss greater than 20% and dilution of original concentration by greater than 50% (Fig. 4). This result shows capping to be a more effective technique for partitioning samples.

Related References

1. Lin, R., D.T. Burke, and M. A. Burns, "Addressable Electric Fields for Size-Fractioned Sample Extraction in Microfluidic Devices," *Analytical Chemistry,* **77,** pp. 4338-4347, 2005.
2. Hung, L.-H., *et al.*, "Alternating Droplet Generation and Controlled Dynamic Droplet Fusion in Microfluidic Device for CdS Nanoparticle Synthesis," *Lab on a Chip,* **6,** pp. 174-178, 2006.
3. Tan, Y.-C., V. Christini, and A. P. Lee, "Monodispersed Microfluidic Droplet Generation by Shear Focusing Microfluidic Device," *Sensors and Actuators B,* **114,** 1, pp. 350-356, 2006.
4. Gartecki, P., *et al.*, "Formation of Droplets and Bubbles in a Microfluidic T-Junction—Scaling and Mechanism of Break-Up," *Lab on a Chip,* **6,** pp. 437-446, 2006.

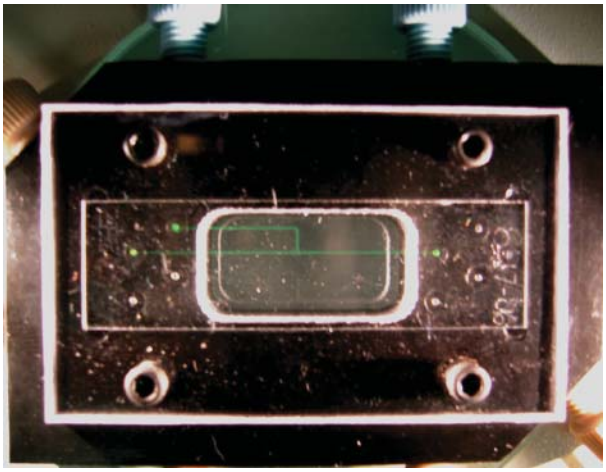

Figure 2. Microfluidic chip and mesoscale package for sample extraction. The extraction protocol can be integrated into existing microfluidic devices with the addition of a single input channel.

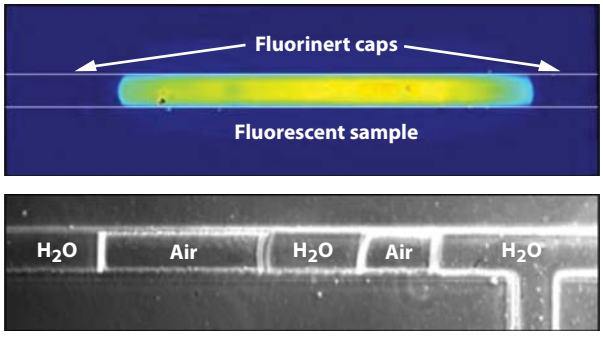

Figure 3. Demonstration of sample capping technique in which air bubbles or fluorinert droplets are injected around a sample. These techniques segregate the sample and enable extraction with minimal loss and dilution.

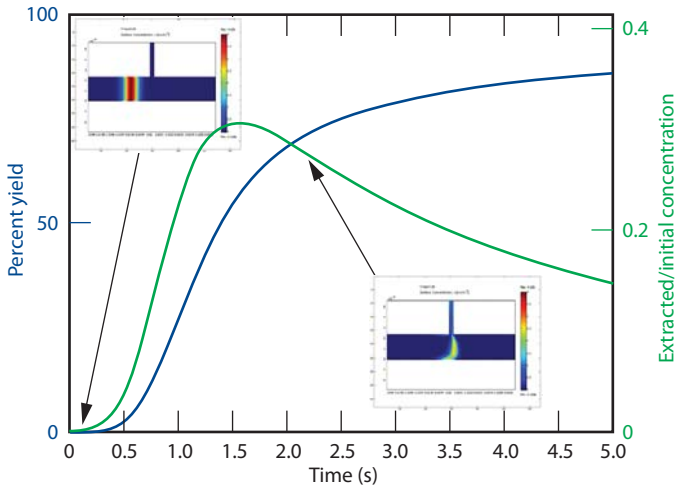

Figure 4. Results from finite element models predicting the extraction yield (blue, left axis) and dilution (green, right axis) for a sample injected into the carrier solution as a function of time. The insets depict the concentration profile of a concentrated plug at times t = 0 s (upper left) and t = 2 s (lower right) as the plug is injected from the primary channel into the side channel with pressure-driven flow. As expected, the percentage of sample extracted increases with time (blue plot) but the concentration (green plot) peaks at ~50% of the sample.